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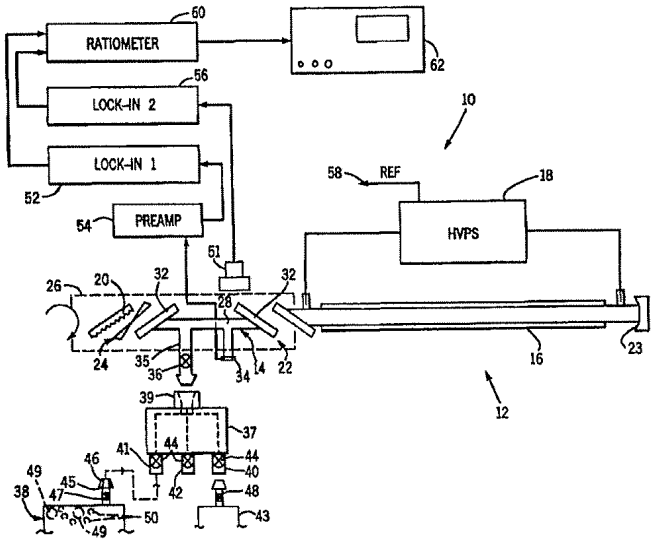

(57) Abstract

A system (10) and method that allows for early detection of biological conditions, such as disease, through analysis of appropriate gaseous samples. The system (10) and method are particularly amenable to the early screening for diseases, such as lung cancer, through the detection of specific biomarkers when present in exhaled breath from an individual or gaseous samples taken proximate cell cultures, pathology specimens, food specimens, etc. The preferred system implements a carbon monoxide laser (16) that generates radiation and directs it through a photoacoustic cell (14). The radiation is of a type that undergoes a characteristic intense absorption by the biomarker, if present, in the gaseous sample. The absorption of the radiation is detected acoustically.

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DESCRIPTION

SYSTEM AND METHOD FOR DETECTION OF A BIOLOGICAL CONDITION

TECHNICAL FIELD

The present invention relates generally to the detection of volatile organic compounds that serve as biomarkers for a biological condition, such as a disease, and particularly to a noninvasive system and method for determining the presence of such condition in an individual or substance through analysis of a gaseous sample from the individual or substance.

BACKGROUND ART

Early detection of disease in an individual is often important to successful treatment of that disease. A variety of techniques are used to test for specific diseases either before or after symptoms occur. For example, blood samples and urine samples are routinely taken for analysis and detection of abnormalities indicative of disease. Many of these techniques are invasive or uncomfortable for the patient.

One potential noninvasive technique for determining the presence of a variety of diseases in the body of an individual is breath analysis. There are over three hundred distinct chemical compounds that may be detected in human expired breath, and each of these distinct chemical compounds has its own absorption spectrum. Studies have shown that specific alterations or changes in this expired air are indicative of specific diseases. This is true for some diseases because of the direct compositional relationship between constituents carried in the blood stream and constituents excreted into the alveolar spaces of the lungs. In any event, the

changes or alterations in the constituents of an individual's breath can be detected to determine whether the individual has a particular biological condition, such as a disease or metabolic disorder.

The presence of a given disease can be indicated by the addition of one or more constituents in a gaseous sample, e.g., expired air, otherwise not present, a change in concentration of one or more constituents or a combination of the two. Thus, a successful diagnostic technique must be able to readily detect the addition or change in concentration of constituents. The addition of a constituent and/or the change in concentration of a constituent are often referred to as biochemical markers or biomarkers. Detection of biomarkers has been a complex task due to the large number of constituents within human expired air and due to the constant change of constituents that results from environmental pollutants and other components which enter an individual's lungs.

To the present, analysis of human expired air has been accomplished by using gas chromatography/mass spectrometry which detects the numerous components within human-expired air. The use of a gas chromatographic/mass spectrometric technique has been necessary because of the number of constituents and because of their presence in sub-microgram per liter to microgram per liter concentrations. Various statistical analyses are performed on the results with the aid of a personal computer to determine the presence of biomarkers. This technique, however, is expensive and time consuming, rendering it inappropriate for the routine testing of individuals.

The gas chromatography/mass spectrometry technique has been used in researching biomarkers associated with lung cancer. Lung cancer is a disease of particular interest due to its growing presence, severe effects and difficulty of early detection. Diagnostic techniques that have been used in the past to detect pre-

symptomatic lung cancer include chest x-rays, fiber-optic bronchoscopy and sputum cytology. However, these techniques are costly to administer and have not been adopted as standard screening techniques. Like the gas chromatography/mass spectrometry approach, these techniques are not amenable to large-scale use for early detection of lung cancer.

Similar problems are involved in detecting other biological conditions. For example, biomarkers contained in the gases resulting from metabolic activity in cell cultures, pathology specimens or food specimens may contain biomarkers indicative of a biological condition, such as disease, spoilage or other contamination.

It would be advantageous to have a quick inexpensive system and method for analyzing gaseous samples, such as the exhaled breath of an individual or the gases proximate cell cultures, pathology specimens, food specimens, etc., to determine detrimental biological conditions, such as spoilage or diseases, e.g., renal failure, liver disease and diabetes, for which specific biomarkers are present in human expired air.

DISCLOSURE OF THE INVENTION

The present invention features a system and method for detecting at least one specific volatile organic compound in a gaseous sample. The specific volatile organic compound is indicative of a biological condition when present in a given amount. The method includes obtaining a gaseous sample that has been exposed to or is suspected of being exposed to byproducts of a biological agent. An electromagnetic radiation generator is selected to generate a radiation that undergoes a characteristic intense absorption by the at least one specific volatile organic compound. The radiation is directed into the sample, and any characteristic intense

absorption is detected. This provides an indication of whether the biological condition exists, based on the presence and amount of the at least one specific volatile organic compound.

According to another aspect of the invention, a system is provided for detecting a biomarker indicative of the presence of a biological condition. The system comprises a photoacoustic cell having an inlet through which a gaseous sample may be introduced into an interior of the photoacoustic cell. An electromagnetic radiation generator is combined with the photoacoustic cell and positioned to direct a radiation through the photoacoustic cell. The selected radiation is of a frequency predetermined for a characteristic intense absorption by at least one specific biomarker contained in the gaseous sample. The system also includes a detector coupled to the photoacoustic cell to detect the characteristic intense absorption indicative of the at least one specific biomarker.

According to another aspect of the invention, a method for detecting whether a given biomarker is present in a selected gaseous sample is disclosed. The method includes obtaining a gaseous sample and selecting a radiation of a type that will undergo a characteristic adsorption by a given biomarker if present in the gaseous sample. The method further includes directing the radiation into the gaseous sample, and detecting whether the characteristic absorption occurs.

BRIEF DESCRIPTION OF DRAWINGS

The invention will hereafter be described with reference to the accompanying drawings, wherein like reference numerals denote like elements, and:

Figure 1 is a schematic illustration of a photoacoustic system according to a preferred embodiment

of the present invention;

Figure 2 is a schematic illustration of a photoacoustic cell that can be used in the system illustrated in Figure 1;

Figure 3 is a representation of an absorption spectrum for one biomarker indicative of lung cancer;

Figure 4 is a representation of another absorption spectrum of a biomarker indicative of lung cancer; and

Figure 5 is a chart showing sample bandwidths and laser lines for a carbon monoxide laser that may be used with the present invention.

BEST MODES FOR CARRYING OUT THE INVENTION

The present invention includes a system and method for detecting biological conditions through noninvasive analysis of gaseous samples. The focus of the following exemplary description is on the detection of biomarkers that are indicative of lung cancer within an individual. However, this system and method can be modified to detect a biomarker(s) in an individual's exhaled breath indicative of other diseases, such as renal failure, liver disease or diabetes. Furthermore , the system and method can be utilized to detect biomarkers expelled into the environment proximate cell cultures, pathology specimens, food specimens, etc., where metabolic processes occur. For example, the system and method can be used for food inspection at a processing facility to insure that the food products have not been contaminated by decay or other infestation of undesirable aerobic or anaerobic organisms. Such organisms undergo metabolic processes that release carbon dioxide or ammonia that can be detected via the present inventive system and method.

Generally, the present system and method utilize photoacoustic detection to determine whether there exist

certain biomarkers within a subject gaseous sample. Biomarkers are individual constituents or combinations of constituents at given concentrations within exhaled breath that indicate a substantial likelihood of the presence of lung cancer (or other biological condition) in the individual or substance being tested.

Examples of biomarkers indicative of lung cancer include ε-caprolactone ($C_6H_{10}O_2$), 2-butanone and acetone. Studies have shown that the presence of ε-caprolactone in the exhaled breath of an individual indicates a ninety percent likelihood that the individual has lung cancer. Similarly, the presence of 2-butanone and the presence of acetone at given concentration levels also provide a strong indication of the presence of lung cancer within the individual. Thus, the presence of these constituents at a given concentration level in the individual's expired air provides a biomarker indicative of lung cancer. The present invention provides a relatively simple, quick and inexpensive system and method for determining the presence of such biomarkers to allow for early screening of lung cancer.

According to a preferred embodiment of the present invention, the pertinent biomarkers can be determined via photoacoustic detection. Generally, a radiation generating source, such as a laser, is used to generate a radiation that may be directed into a gaseous sample, e.g., exhaled breath from a subject or substance being tested. Preferably, the laser is a carbon monoxide laser tuned to a selected wavelength or wavelengths. The biomarkers of interest are known to have infrared absorption spectra, and therefore the wavelength or wavelengths of the radiation is selected from within a specific band of infrared wavelengths.

In testing for many types of disease, a breath sample is placed inside a photoacoustic cell, and the radiation is directed through the cell. A microphone is connected to the photoacoustic cell and exposed to the

breath sample therein. The microphone is able to detect absorption of the radiation by a specific biomarker via pressure changes within the photoacoustic cell. For example, the laser may be tuned to generate radiation having a wavenumber of approximately 1768 cm^{-1} which corresponds to a characteristic intense absorption, e.g., an absorption peak, of ε-caprolactone.

Preferably, the radiation from the laser is chopped at a certain frequency, thereby producing a corresponding modulation of absorption of the radiation by the sample of breath within the photoacoustic cell. Assuming the appropriate biomarker is present, the absorption of radiation by the biomarker results in a heating of the biomarker gas that generates an acoustic wave detectable by the microphone. An acoustic signal enhancer, such as helium, can be added to the sample of breath being tested to facilitate generation of this acoustic wave.

Referring now to Figure 1, a photoacoustic detection system 10, according to a preferred embodiment of the present invention, is schematically illustrated. Although this is a preferred embodiment, a variety of component arrangements and modifications may be made without departing from the scope of this invention. In the illustrated embodiment, an electromagnetic radiation generator or radiation source 12 is used to generate a desired radiation having a wavelength or wavelengths that will be characteristically absorbed by a specific biomarker or biomarkers, if present within the gaseous sample being analyzed. The sample is contained within a photoacoustic cell 14 during the analysis.

In the preferred embodiment, radiation source 12 is a continuous wave, grating-tuned carbon monoxide (CO) laser 16 connected to a high voltage power supply 18. Laser 16 operates on one or more selected transitions of $^{12}C^{16}O$ and/or other isotopic variants of carbon monoxide depending on a given biomarker's absorption spectrum and

the location of its characteristic intense absorption within that spectrum. The laser transitions are tuned by a diffraction grating 20 that is typically mounted on a rotation stage (not shown) as is understood by those of ordinary skill in the art. The laser 16 is modulated either by voltage modulation of its high voltage power supply 18 or by a mechanical chopper 24, preferably disposed with an optical cavity 22. Optical cavity 22 is located between a cavity mirror 23 of laser 16 and diffraction grating 20.

Although photoacoustic cell 14 can be disposed outside optical cavity 22, it preferably is located within optical cavity 22 to use the intracavity power of the individual laser lines. Correspondingly, this intracavity placement of the photoacoustic cell will increase the detection sensitivity of photoacoustic detection system 10, often by a factor of 100 or more. In the illustrated embodiment, a purge box 26 (shown by dashed lines) encloses photoacoustic cell 14, diffraction grating 20, chopper 24 (if used) and the end of laser 16 proximate photoacoustic cell 14.

Photoacoustic cell 14 can be designed in a variety of configurations to optimize its effectiveness. A preferred exemplary embodiment is illustrated in Figures 1 and 2. In this embodiment, photoacoustic cell 14 includes a generally linear body 28 that lies along an axis 30 (see Figure 2). Linear body 28 preferably has a hollow interior 31 that is generally cylindrical in cross section. A pair of cell windows 32, such as ZnSe windows, are mounted to linear body 28 with one window 32 at each end. Preferably, cell windows 32 are each mounted at Brewster's angle to reduce intracavity losses. Additionally, a microphone 34, such as an electret microphone, is coupled to photoacoustic cell 14 and exposed to its interior cavity 31 as well as any samples of breath introduced therein. An exemplary microphone is a Knowles Electronics BT-1759 electret microphone.

An inlet/outlet port 35 is connected to linear body 28 to permit the gaseous sample, e.g., expired breath, to be introduced into interior cavity 31 and removed therefrom. Additionally, it may be desirable to introduce an acoustic signal enhancer, such as helium, into interior cavity 31. Additionally, it may also be desirable to introduce a calibration gas, such as SF_6, into interior cavity 31. Potentially, both the gaseous sample and helium can be introduced into photoacoustic cell 14 through port 35.

Port 35 includes a valve 36 that allows the ingress of the gaseous sample (and, if used, the acoustic signal enhancer) but not its egress during analysis. Following analysis of the sample, valve 36 is opened and the sample is extracted or sucked from interior cavity 31. After removal of the sample, valve 36 is shut to retain a vacuum within interior cavity 31. This lower pressure allows the next gaseous sample to be expanded into photoacoustic cell 14 via the higher outside pressure that forces the sample into the lower pressure or vacuum within interior cavity 31.

In the illustrated embodiment, port 35 is designed for coupling to a manifold 37 which, in turn, is designed for coupling to a sample container 38. Sample container 38 may comprise a variety of configurations, such as a preevacuated flexible bag that can be filled with the desired gaseous sample within a sample collection container used to collect exhaled breath or the gases proximate other biological agents, e.g., cell cultures, pathology specimens or food specimens, that potentially have been exposed to byproducts of the metabolism of the biological agent. In the case of breath analysis, the individual (biological agent) potentially can exhale directly into the flexible bag to provide the sample for analysis. The container 38 preferably includes a valved port to permit entry of the sample and prevent any leakage

or unwanted contamination.

Manifold 37 includes a connection port 39, designed for mating engagement with port 35. Manifold 37 also includes a helium port 40, a sample port 41 and an evacuation port 42. A helium supply 43 (see Figure 2) may be attached to helium port 40; sample container 38 may be attached to sample port 41; and a vacuum pump (not shown) may be attached to evacuation port 42. A valve 44 is disposed in each port 40 and 41 to permit inflow of the helium and the breath sample. A similar valve 44 is disposed in evacuation port 42 to facilitate evacuation of manifold 37, interior cavity 31 and the "dead space" between valves.

Sample container 38 includes an outlet port 45 designed for sealing engagement with sample port 41 of manifold 37. Outlet port 45 can also serve as the port through which the sample is admitted into container 38, or a separate inlet port can be provided depending on the specific application. In one embodiment, outlet port 45 includes a contoured surface 46, such as a tapered octagonal or hexagonal surface that is designed for mating engagement with a corresponding contoured surface of sample port 41. This will help ensure that only the desired sample container 38 is connected to manifold 37 and ultimately photoacoustic cell 14. After making the connections, the sample of breath can be expanded or otherwise forced into interior cavity 31 of photoacoustic cell 14. In lieu of a single inlet/outlet port 35, photoacoustic cell potentially can be designed with separate inlet and outlet ports connected to body 28.

In the preferred embodiment, sample container 38 includes a valve 47 disposed in outlet port 45. Similarly, the helium supply 43 includes a valve 48. This permits a sample of breath to be introduced into interior cavity 31 of photoacoustic cell 14 as follows: Manifold 37 is connected to port 35 of photoacoustic cell 14 via connection port 39. Sample container 38, with valve 47

closed, is connected to sample port 41 via outlet port 45, helium supply 43, with valve 48 closed, is connected to helium port 40, and a vacuum is applied at evacuation port 42. Valves 47 and 48 remain closed, and valve 36 as well as valves 44 are opened to evacuate interior cavity 31 and manifold 37 via the vacuum applied at evacuation port 42.

After evacuation, valve 44 of evacuation port 42 is closed, and valve 47 of sample container 38 is opened to permit expansion of the gaseous sample into interior cavity 31 of photoacoustic cell 14. Valve 47 may then be closed and valve 48 opened to permit the flow of helium into interior cavity 31. Generally, the helium is at a higher pressure than the gaseous sample, so it will readily flow into photoacoustic cell 14. A pressure gauge (not shown) may be connected to manifold 37 for monitoring both the sample pressure and the helium pressure to facilitate preparation of reproducible and/or optimized mixtures in interior cavity 31. Of course, the exact method for handling and testing the gaseous sample may vary depending on the type of photoacoustic cell and/or manifold that is used and whether an acoustic signal enhancer, such as helium, and/or a calibration gas, such as SF_6, is combined with the sample.

Further, it may be advantageous to remove water vapor from the sample of breath prior to analysis. Water vapor removal can be accomplished while the sample is contained in sample container 38 by including a desiccant 49 within the sample container. It may also be advantageous to remove other major constituents of expired breath, such as nitrogen, oxygen and carbon dioxide. This can be accomplished by including an appropriate adsorber 50 for the removal of one or more of these other constituents that are unnecessary for analysis of the gaseous sample but may reduce the photoacoustic signal level.

Microphone 34 is designed to detect pressure

changes within photoacoustic cell 14. The pressure changes result from the heating of specific gaseous constituents, i.e., biomarkers, that may be present in the sample of breath undergoing an analysis within interior cavity 31. These pressure changes in photoacoustic cell 14 result from the characteristic intense absorption by one or more of the biomarkers. Typically, a specific biomarker's characteristic intense absorption is a substantially increased absorption of the radiation generated at a specific frequency by laser 16 and directed through photoacoustic cell 14.

However, even when a biomarker indicative of lung cancer is present in the sample being analyzed, only a small portion of the radiation emitted from laser 16 is absorbed within photoacoustic cell 14. A photodetector 51 is employed to monitor the power of laser 16 within photoacoustic cell 14. Photodetector 51 is used to help compensate for changes detected by microphone 34 due to changes in the power of radiation generated by laser 16.

In the embodiment illustrated, microphone 34 is coupled to an amplifier 52 preferably via a preamplifier 54 which receives the signal provided by microphone 34. Similarly, photodetector 51 is coupled to and provides a signal to an amplifier 56. The preamplifier 54/amplifier 52 combination detects and conditions microphone signals at the selected modulation frequency which optimizes the signal-to-noise ratio. Amplifiers 52 and 56 are preferably lock-in amplifiers which provide synchronous detection and reduce the influence of extraneous effects, such as noise, on the measurement of both the pressure changes within photoacoustic cell 14 and the radiation generated by laser 16.

Also, the modulation of the radiation emitted by laser 16 is synchronized with the gating of amplifiers 52 and 56. This may be accomplished by providing a reference signal 58 from the modulation source (e.g., chopper 24, if used, or voltage modulated high voltage power supply 18).

Reference signal 58 is directed to lock-in amplifiers 52 and 56 which are able to detect signals that are synchronous with the modulation (i.e., at the same modulation frequency, with or without a phase shift).

The amplified signals from amplifiers 52, 56 are directed to a ratiometer 60 that, in turn, provides an output signal which is effectively a measure of the ratio of the signal detected by microphone 34 divided by the signal detected by photodetector 51. In other words, the signal is a measure of the fraction of the total input radiation that is absorbed by a given biomarker or biomarkers within photoacoustic cell 14. This signal may then be directed to a data acquisition, analysis and display system 62 that is able to record, process, analyze and/or display the ratioed photoacoustic signal, as would be understood by one of ordinary skill in the art. It should be noted that the signal provided by microphone 34 can be further enhanced by introducing an acoustic modifier, such as helium, into photoacoustic cell 14 along with the sample being analyzed.

The above described system provides very sensitive measurements, and has the ability to detect the presence of constituents that may only appear within the sample in parts per billion by volume. However, photoacoustic detection system 10 also can accurately detect the concentration levels of constituents within the gaseous sample. This is important for some constituents where changes in concentration from a normal level serve as a biomarker for the presence of a biological condition, such as lung cancer or other disease.

The concentration or density of a specific constituent is determined by measuring the signal strength of the signal detected by microphone 34 and comparing this to the signal strength(s) of a reference sample or samples of known concentration(s), as is understood in the art. Photoacoustic cell 14 is first calibrated using a reference sample or samples to measure signal levels

provided by microphone 34 for known concentrations of the subject constituent. Over a broad range of concentrations, the photoacoustic signal level is linearly related to the concentration of a given dilute constituent within a sample mixture containing the constituent plus the acoustic signal enhancer, if used, all at a fixed total pressure. The concentration or density of the subject constituent within the sample can then accurately and readily be determined based on comparing the signal strength of the acoustic wave created from the constituent's characteristic absorption to those of the reference sample or samples.

One exemplary implementation of the present invention provides a method for detecting lung cancer in an individual, and includes analysis of a sample of the individual's breath to detect one or more lung cancer biomarkers. The sample may, for example, be originally provided by the individual via blowing into a sample container, such as sample container 38. The sample then can be analyzed or sent, e.g., mailed, to a central processing location where it is obtained for analysis.

According to further aspects of the invention, a laser or other appropriate radiation source is selected to generate a radiation that undergoes the characteristic intense absorption by the biomarker of interest. The radiation is directed into the sample of breath, and a detector, such as microphone 34, detects whether the characteristic absorption of the radiation occurs.

Referring generally to Figures 3-4, representative absorption spectra of specific biomarkers indicative of lung cancer are illustrated. In Figure 3, for example, a representative absorption spectrum for a lactone, specifically ε-caprolactone, shows the absorbance of this biomarker versus the wavenumber of the radiation for a particular concentration and path length of this chemical compound. As illustrated, a characteristic intense absorption 64 occurs when the constituent is

struck by radiation having a wavenumber of approximately 1768 cm^{-1}. Assuming there is no interference, i.e., overlapping absorption spikes, from other constituents within the sample of breath, laser 16 can be tuned to emit radiation with a wavenumber, e.g., approximately 1768 cm^{-1}, to match the characteristic intense absorption 64 of the biomarker ε-caprolactone. This characteristic absorption within photoacoustic cell 14 creates a pressure pulse detected by microphone 34. As described above, microphone 34 provides a signal that is ultimately displayed on unit 62 to indicate the presence of biomarker ε-caprolactone and the consequent probability of lung cancer in the individual from which the sample of breath originated.

A biomarker, such as ε-caprolactone, is normally a constituent in exhaled breath only when the individual has lung cancer. Thus, detecting its mere presence in the sample of breath is a strong biomarker indicative of lung cancer. Additionally, there appears to be little if any interference between the characteristic intense absorption of ε-caprolactone and that of other constituents within exhaled breath, provided the water vapor is removed from the breath sample. Thus, photoacoustic detection system 10 can be used to simply detect the presence of this constituent within the sample of breath to provide an indication of lung cancer.

Other constituents, such as acetone, may appear in the expired air of individuals without lung cancer. However, this constituent still provides an indication or a biomarker of lung cancer when it appears at certain densities or concentrations within the exhaled breath of the test subject. The concentration of acetone can readily be determined, as described above, once the photoacoustic signal level is properly calibrated using a reference mixture or mixtures, as known by those of ordinary skill in the art.

As illustrated in Figure 4, acetone undergoes a characteristic intense absorption 66 in the presence of

radiation having a wavenumber of approximately 1740 cm^{-1}. Thus, when detecting acetone, laser 16 is tuned to emit radiation having a wavenumber of approximately 1740 cm^{-1}. Again, this causes the characteristic absorption within the sample and creates an acoustic wave within photoacoustic cell 14 that is detected by microphone 34. With a properly calibrated photoacoustic cell 14, the signal strength can be compared to the signal strength of a reference sample of known concentration to determine the density of acetone within the sample. At certain densities, the acetone serves as a biomarker indicative of lung cancer within the individual.

It should be noted that the optimum characteristic intense absorption may not be the maximum absorption because of interference at that wavenumber. For example, if the subject biomarker and another biomarker or constituent both absorb radiation at a given wavenumber, it may be desirable to use another wavenumber to optimize detection of the desired biomarker. Regardless of the specific wavenumber at which a given biomarker undergoes the desired characteristic absorption, laser 16 can be finely tuned to emit radiation having the desired wavenumber. In fact, it is often desirable to detect the presence, and possibly the concentration, of more than one biomarker. This may be accomplished by appropriately tuning laser 16 to emit the desired radiation for each biomarker sought.

Preferably, laser 16 is a carbon monoxide laser having great flexibility with respect to control over the wavenumber at which radiation is emitted. This concept may be clarified with reference to Figure 5 which includes a chart showing the relative laser gain coefficients for individual rotational lines within lasing vibrational bands V=15→14 through V=10→9 of carbon monoxide. As illustrated by the chart, selection of the appropriate vibrational band and rotational line or lines within that

band permits great control over the wavenumber of radiation emitted from laser 16. The illustrative chart in Figure 5 is based on the $^{12}C^{16}O$ isotope of carbon monoxide, but numerous adjustments to the position of the laser lines within a given vibrational band, and consequently the wavenumber of the radiation emitted, can be made by choosing other isotopic variants of carbon monoxide. This permits laser 16 to be accurately tuned to emit radiation having a desired wavenumber for absorption by a biomarker even if the characteristic intense absorption of that biomarker is spread over a small range of wavenumbers or fractions of wavenumbers.

With certain diseases or other biological conditions, there may be interference between the absorption spectra of biomarkers and other constituents within the gaseous sample. Sometimes, the interfering constituents can be removed from the sample prior to analysis. As described above, water vapor, which tends to show substantial absorption at a wide range of wavenumbers, can be removed by an appropriate desiccant. Similarly, other constituents can be removed, prior to analysis, with appropriate adsorbents. However, the interfering constituents are not always amenable to early removal.

In these situations, it may be necessary to determine the photoacoustic signal contributions of each of the interfering gases within the sample. The gases of interest will tend to have interfering absorption spectra over a definite range of wavenumbers, and the actual gases involved can be determined from the various known absorption spectra of the constituents found in exhaled breath or other gaseous sample.

To determine the photoacoustic signal contributions of the interfering gases, laser 16 is adjusted to emit radiation having a predetermined wavenumber. The radiation is directed through photoacoustic cell 14 and the photoacoustic signal is

measured. This procedure is repeated at different predetermined wavenumbers. The number of wavenumbers should be at least one greater than the sum of the number of biomarkers plus the number of interfering gases whose photoacoustic signal contributions are of interest. Preferably, the predetermined wavenumbers of the radiation are chosen for strong absorption by each of the different gaseous constituents of interest. The overall photoacoustic signal measured for each radiation having a given wavenumber can be treated as the linear sum of photoacoustic signal contributions by each relevant constituent of the sample. This can be described in algebraic form as follows:

$$S_j = \sum_{i=1}^{n} s_{ij}$$

S_j = the total photoacoustic signal provided by the mixture at a radiation having a given wavenumber j

n = the number of constituents i in the mixture

s_{ij} = the photoacoustic signal contribution for a particular constituent i, i.e. gas, at a particular wavenumber j

Each photoacoustic signal contribution s_{ij} of constituent i at wavenumber j is the product of a concentration for that constituent c_i times its signal contribution at wavenumber j per unit concentration σ_{ij}:

$$s_{ij} = c_i \sigma_{ij}$$

The quantity σ_{ij} is termed the "normalized photoacoustic signal contribution". The normalized photoacoustic signal contribution for each constituent i at each wavenumber j can be determined by measuring photoacoustic spectra (i.e., photoacoustic signals as a function of wavenumber)

of reference samples of each constituent.

The concentrations of each of the constituents c_i can be determined by, for example, least squares minimization of the residuals of calculated *vs.* measured total photoacoustic signal measurements S_j as a function of trial values c_i. The calculations are typically performed on a processor or computer incorporated into unit 62 of photoacoustic system 10. By this method, the density or concentration of the constituents can be used to determine whether a biomarker indicative of a particular disease is present. If multiple interfering biomarkers are potentially present, this type of analysis also can be used to determine the concentrations of those multiple biomarkers.

Another exemplary procedure involves the use of a continuous wave (cw) CO laser operating in the 2.6 to 4.0 μm spectral region. This procedure utilizes cw CO laser transitions that occur in the 2.6 to 4.0 μm spectral region based on the $\Delta v = -2$ vibrational band sequence, such as the $v=13\rightarrow11$, $12\rightarrow10$, etc. vibrational bands. Use of this spectral region helps avoid spectral interference from water vapor and other contaminants in determining VOCs present in the gaseous sample, e.g., breath sample, cell culture sample, food specimen sample, etc.

It will be understood that the foregoing description is of preferred exemplary embodiments of this invention and that the invention is not limited to the specific form shown. For example, a variety of photoacoustic cell designs may be used; a variety of data acquisition, processing and/or display units may be incorporated into the system; and the overall system may be adapted to screen for various biomarkers indicative of a wide variety of biological conditions. These and other modifications may be made in the design and arrangement of the elements without departing from the scope of the invention as expressed in the appended claims.

CLAIMS

What is claimed is:

1. A method for detecting at least one specific volatile organic compound in a gaseous sample, the at least one specific volatile organic compound being indicative of a biological condition when present in a given amount, the method including obtaining a gaseous sample that has been exposed to or is suspected of being exposed to byproducts of a biological agent, characterized by:

selecting an electromagnetic radiation generator (12) able to generate a radiation that undergoes a characteristic intense absorption (64, 66) by the at least one specific volatile organic compound;

directing the radiation into the sample; and

detecting whether the characteristic intense absorption (64, 66) of the radiation occurs to thereby provide an indication of whether the biological condition exists, based on the presence and amount of the at least one specific volatile organic compound.

2. The method as recited in claim 1, further characterized in that the step of detecting includes detecting an acoustic wave.

3. The method as recited in claim 1, further characterized in that the step of detecting includes the step of implementing photoacoustic detection to determine the presence and amount of the at least one specific volatile organic compound.

4. The method as recited in claim 1, further characterized by the step of combining the gaseous sample

with an acoustic signal enhancer.

5. The method as recited in claim 1, further characterized in that the step of selecting includes selecting a laser (16) able to generate radiation that undergoes the characteristic intense absorption (64, 66).

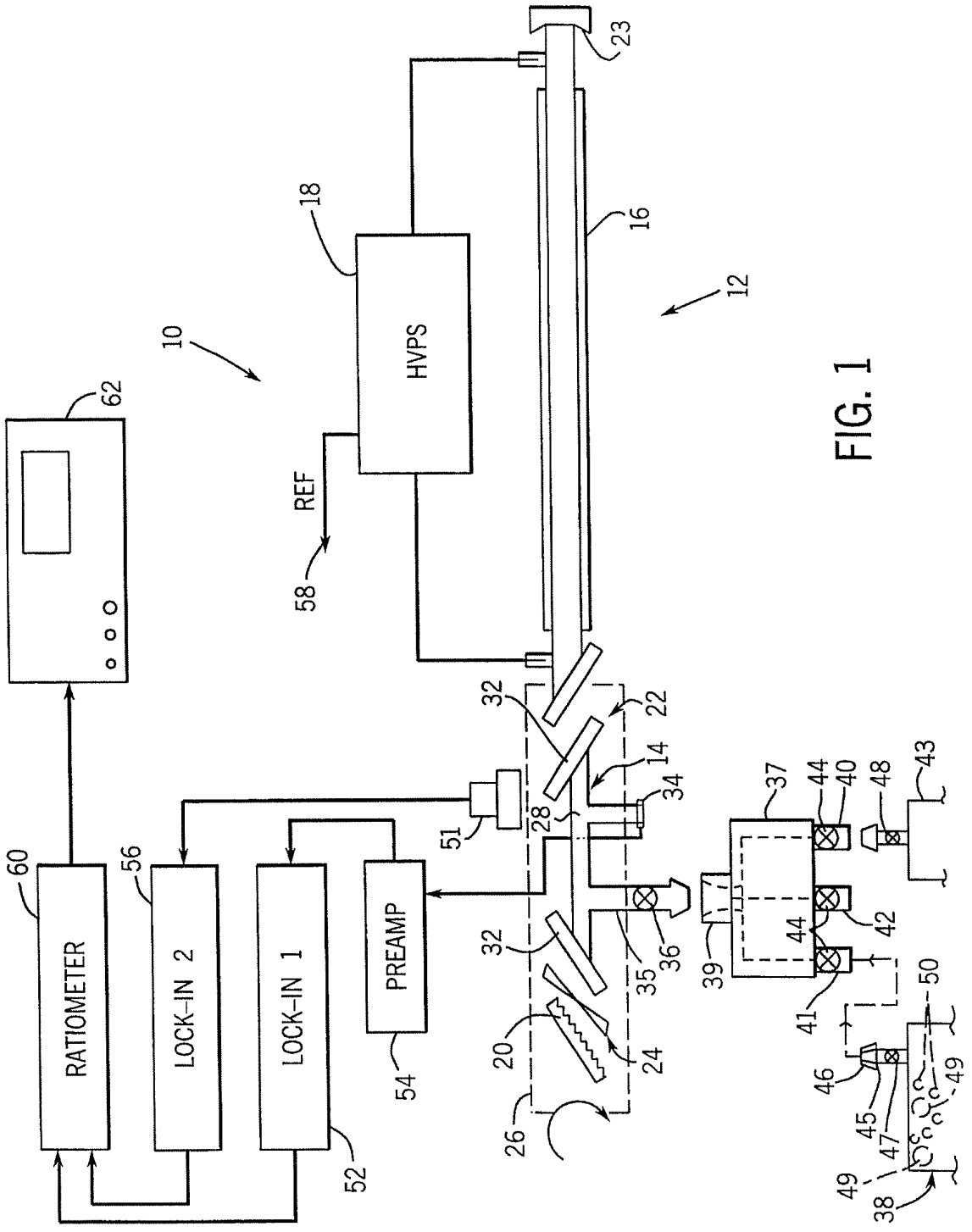
6. The method as recited in claim 5, further characterized by the step of placing the gaseous sample within an optical cavity (22) of the laser (16).

7. The method as recited in claim 5, further characterized in that the step of selecting includes selecting a laser (16) able to generate radiation having a wavenumber within the range from 1600 cm^{-1} to 1900 cm^{-1}.

8. The method as recited in claim 1, further characterized in that the step of selecting includes selecting a carbon monoxide laser (16).

9. The system as recited in claim 5, further characterized by the step of placing the gaseous sample into a photoacoustic cell (14) to enhance detection of the characteristic intense absorption (64, 66) when the radiation is directed into the sample.

10. The system as recited in claim 9, further characterized by the step of placing the photoacoustic cell (14) in an optical cavity (22) of the laser (16).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LOCK-IN 1
PREAMP
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FIG. 1

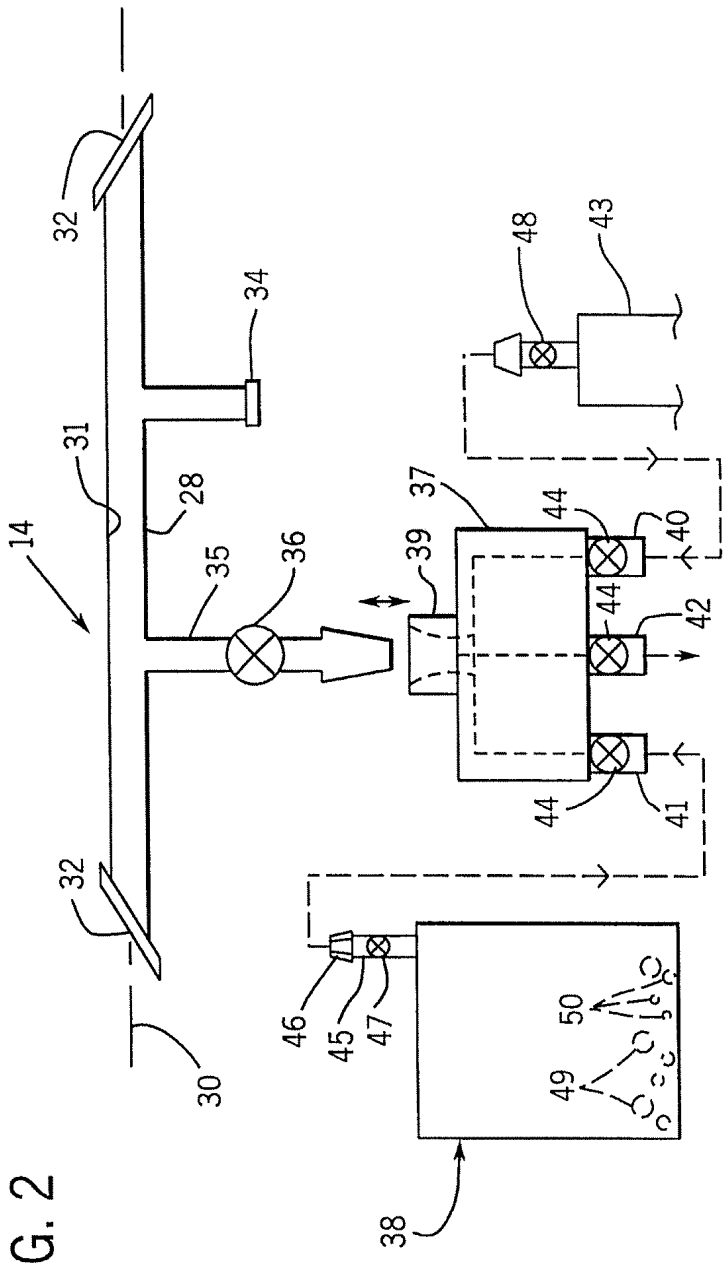
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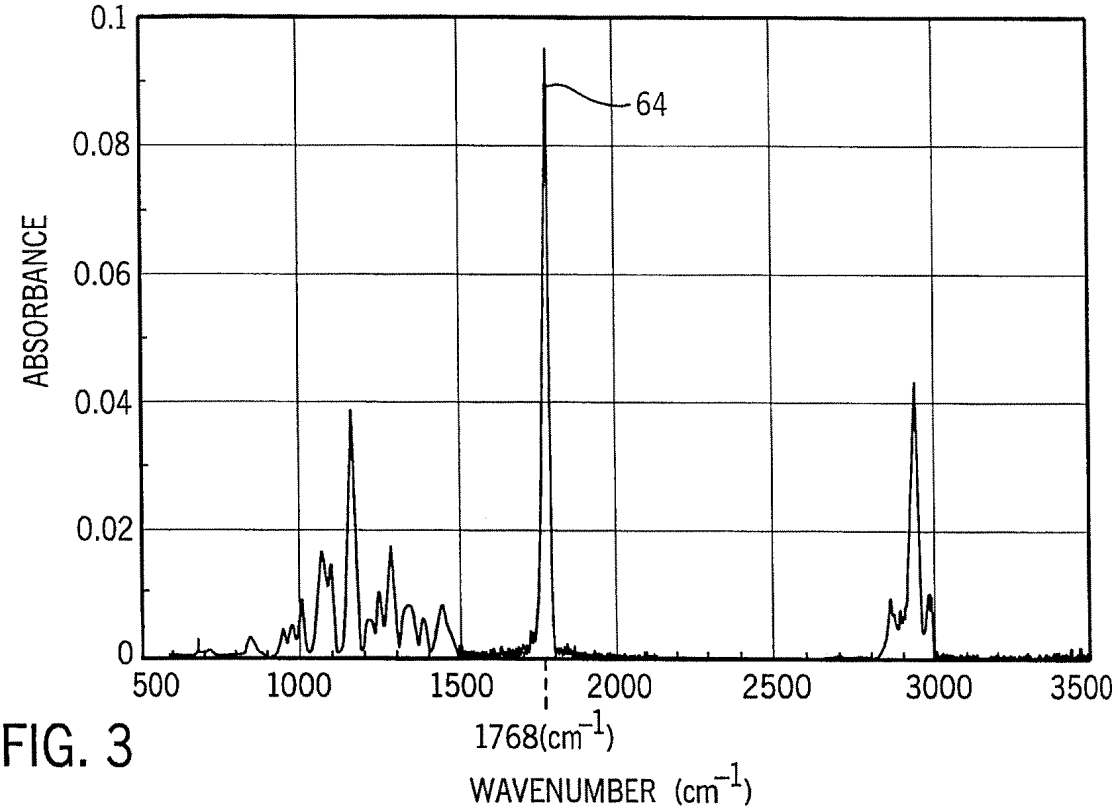
FIG. 2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0.08
0.06
0.04
0.02
0
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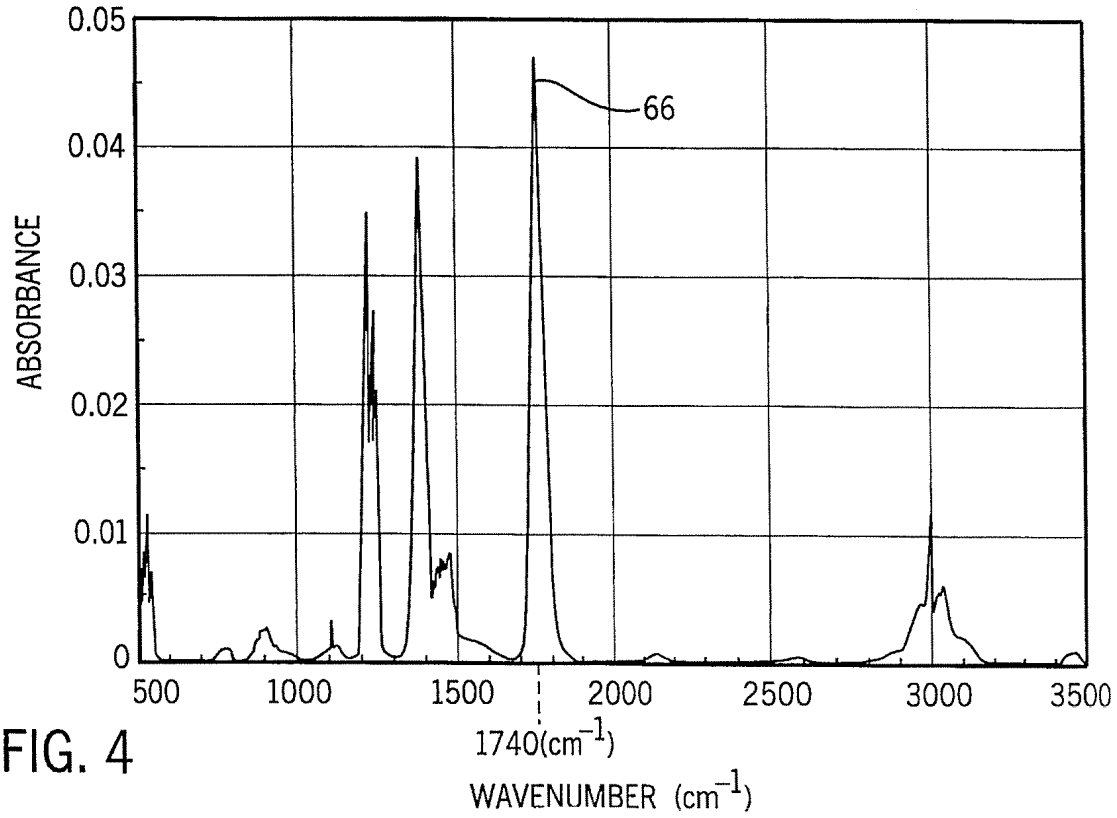
1500
2000
2500
3000
3500
1768(cm-1)
WAVENUMBER (cm-1)

FIG. 3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0.02
0.01
0
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1000
1500
2000
2500
3000
3500
1740(cm-1)
WAVENUMBER (cm-1)

FIG. 4

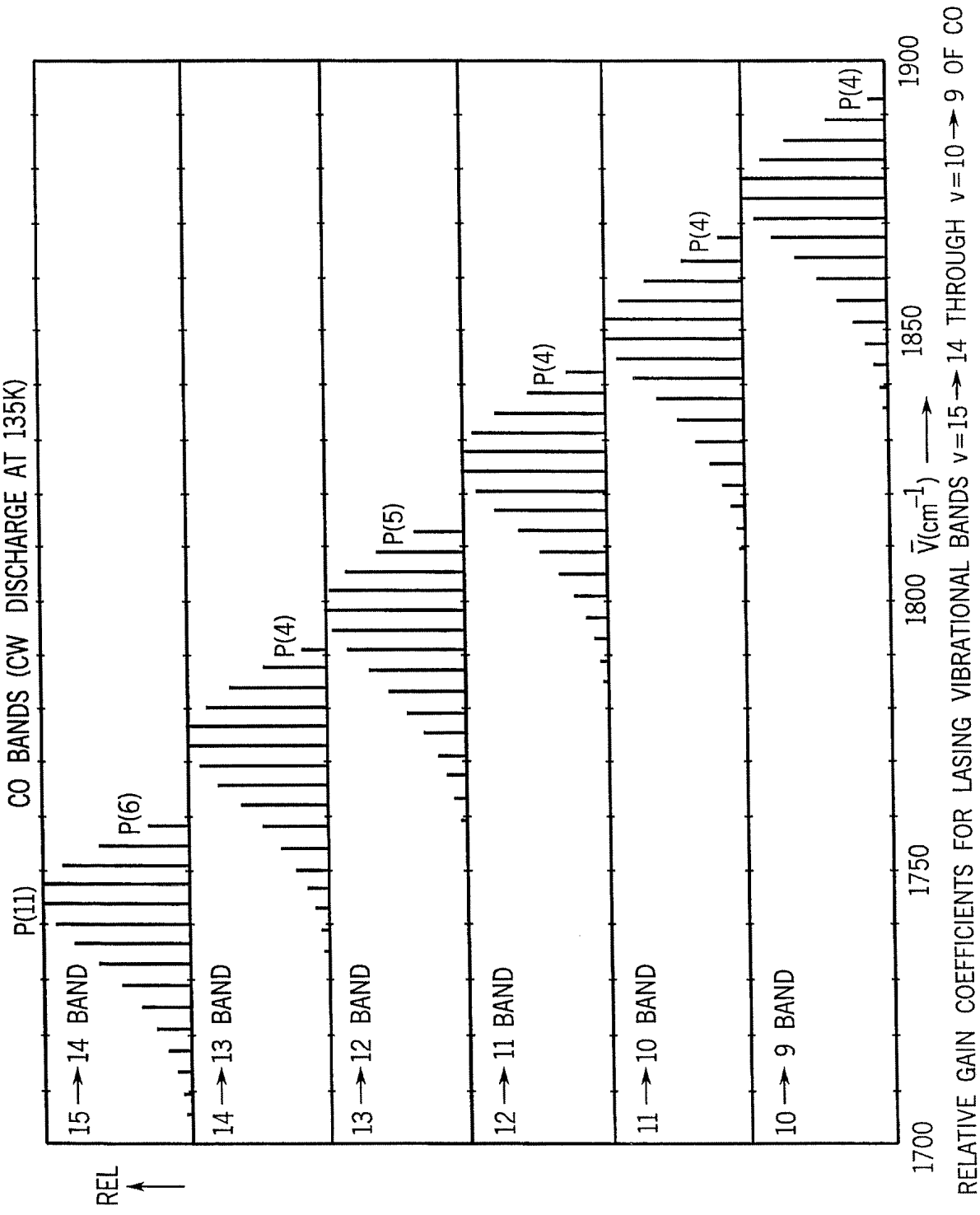

FIG. 5

RELATIVE GAIN COEFFICIENTS FOR LASING VIBRATIONAL BANDS $v=15 \rightarrow 14$ THROUGH $v=10 \rightarrow 9$ OF CO

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US98/10771

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) :G01N 21/00, 33/497
US CL :436/148, 164, 165, 181, 900; 422/82.05, 82.09, 83, 84
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 436/128, 130, 148, 164, 165, 181, 900; 422/82.05, 82.09, 83, 84; 356/51, 301

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

APS, STN/CA, BIOSIS, MEDLINE, WPID
search terms: lung, cancer, breath, acoustic, photoacoustic, radiation

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	O'NEILL, H.J. et al. A Computerized Classification Technique for Screening for the Presence of Breath Biomarkers in Lung Cancer. Clinical Chemistry. 1988, Vol. 34, No. 8, pages 1613-1618, especially page 1613.	1-10
Y	Chemical Abstracts, AN 126:128980, DE 19522774 A1, SCHARFF, W. et al. Apparatus for Spectroscopic Investigation of Samples from the Human Body. 02 Januray 1997.	1-10
Y	BERNEGGER, S. et al. Longitudinal Resonant Spectrophone for CO-Laser Photoacoustic Spectroscopy. Applied Physics. 1987, B44, pages 125-132, especially page 129.	1-10

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

23 JULY 1998

Date of mailing of the international search report

01 SEP 1998

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